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31.03.93 Bulletin 93/13(71) Applicant: **EXOEMIS, INC.**
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Kilburn & Strode 30, John Street
London WC1N 2DD (GB)(54) **Haloperoxidase-acid-optimum chemiluminescence assay system.**

(57) A highly sensitive chemiluminescent indicator system is disclosed for determining the presence or amount of an analyte in a liquid sample. The new acid optimum chemiluminescent indicator system comprises haloperoxidase (halide:hydrogen peroxide oxidoreductase), halide, oxidant and chemiluminogenic substrate. The indicator system acts as a synthesizer of highly reactive singlet molecular oxygen (¹O₂), which reacts with the chemiluminogenic substrate to yield an excited state, oxidized reaction product. The excited state reaction product then relaxes to a lower energy (e.g., ground) state with the emission of measurable light in an amount related to the amount of each of the reaction participants present in a reaction solution. Known, non-rate limiting amounts of three of the reaction participants are provided in an assay solution to determine the presence or amount of the fourth participant in a test sample. The fourth participant in the test sample may be the analyte of interest, or may be produced or consumed in the test sample through one or more preliminary reactions involving the ultimate analyte of interest, with the amount of the fourth participant being related to the amount of analyte in the test solution. Accordingly, the indicator system of the invention may be employed for the determination of a wide variety of analytes. The indicator system operates most efficiently over the range from acid to slightly basic pH, e.g., at a pH of about 3 to about 8. Also described is a variety of

illustrative assay formats in which the indicator reaction of the invention may be employed, as well as kits for use in carrying out assays utilizing the haloperoxidase/halide/oxidant/substrate indicator system.

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EUROPEAN SEARCH REPORT

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DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl.5)
A	BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS vol. 63, no. 3, 1975, NEW YORK NY USA pages 684 - 691 R.C. ALLEN 'The role of pH in the chemiluminescent response of the myeloperoxidase-ide-HOOH antimicrobial system.' * the whole document * ---	1-23	C12Q1/28 G01N33/84
A	BIOCHEMISTRY vol. 26, 1987, WASHINGTON DC USA pages 5127 - 5132 V. RENGANATHAN ET AL. 'Haloperoxidase reations catalyzed by lignin peroxidase, an extracellular enzyme from the Basidiomycete Phanerochaete chrysosporium.' * the whole document * -----	1-23	TECHNICAL FIELDS SEARCHED (Int. Cl.5) C12Q G01N
The present search report has been drawn up for all claims			
Place of search THE HAGUE		Date of completion of the search 22 JANUARY 1993	Examiner VAN BOHEMEN C.G.
CATEGORY OF CITED DOCUMENTS X : particularly relevant if taken alone Y : particularly relevant if combined with another document of the same category A : technological background O : non-written disclosure P : intermediate document T : theory or principle underlying the invention E : earlier patent document, but published on, or after the filing date D : document cited in the application L : document cited for other reasons & : member of the same patent family, corresponding document			